Datasheet for 600-401-A77

ABCB5 Antibody

Overview

Description:	Anti-ABCB5 (RABBIT) Antibody - 600-401-A77
Item No.:	600-401-A77
Size:	100 µg
Applications:	ELISA, IHC, WB, FC
Reactivity:	Human
Host Species:	Rabbit

Product Details

Background:	This antibody is designed, produced, and validated as part of a collaboration between Rockland and the National Cancer Institute (NCI) and is suitable for Cancer, Immunology and Nuclear Signaling research. Anti-ABCB5 Antibody is a ATP-Binding Cassette protein. ABCB5 is a novel member of the human P-glycoprotein family. It functions as a determinant of membrane potential and regulator of cell fusion in physiologic skin progenitor cells. Cell fusion is thought to contribute to tissue differentiation. ABCB5 is predominantly expressed in melanoma cells and is a novel molecular marker for a distinct subset of chemoresistant stem cell phenotype-expressing tumor cells among human epidermal melanocytes. ABCB5 is a rhodamine efflux transporter. This antibody product is intended to be used to confirm cellular localization and expression level of ABCB5. ABCB5 antibody is ideal for Cancer and Signal Transduction research.
Synonyms:	rabbit anti-ABCB5 Antibody, ABCB 5, ABCB-5, ATP binding cassette sub family B (MDR/TAP) member 5 antibody, ATP binding cassette sub family B member 5 antibody, EST422562 antibody, P glycoprotein ABCB5 antibody
Host Species:	Rabbit
Clonality:	Polyclonal
Format:	IgG

Target Details

Gene Name:	ABCB5
Reactivity:	Human

Immunogen Type:	Conjugated Peptide
Immunogen:	ABCB5 antibody was prepared from whole rabbit serum produced by repeated immunizations with a synthetic peptide corresponding to residues corresponding to an internal region of human ABCB5.
Purity/Specificity:	This product was affinity purified from monospecific antiserum by immunoaffinity chromatography. This antibody is specific for human ABCB5 protein. A BLAST analysis was used to suggest partial cross-reactivity with ABCB5 from monkey (85% homology), rat (68% homology) and mouse (62% homology) sources. Cross-reactivity with ABCB5 from other sources has not been determined.
Relevant Links:	• NCBI - 56849536 • UniProtKB - Q2M3G0 • GeneID - 340273

Application Details

Tested Applications:	ELISA, IHC, WB
Suggested Applications:	FC (Based on references)
Application Note:	This affinity purified ABCB5 antibody has been tested for use in ELISA, IHC, and western blotting. Specific conditions for reactivity should be optimized by the end user.
Assay Dilutions:	All assays should be optimized by the user. Recommended dilutions (if any) may be listed below.
ELISA:	1:10,000 - 1:50,000
IHC:	1:200
WB:	1:20,000

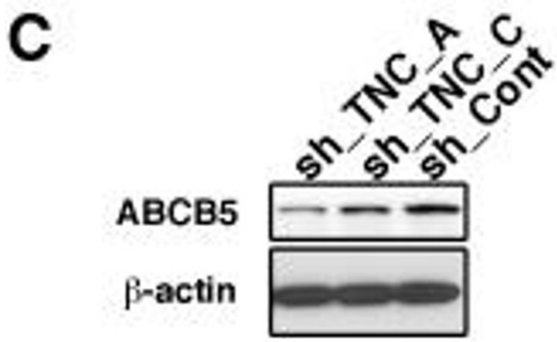
Formulation

Physical State:	Liquid (sterile filtered)
Concentration:	1.1 mg/mL by UV absorbance at 280 nm
Buffer:	0.02 M Potassium Phosphate, 0.15 M Sodium Chloride, pH 7.2
Preservative:	0.01% (w/v) Sodium Azide
Stabilizer:	None

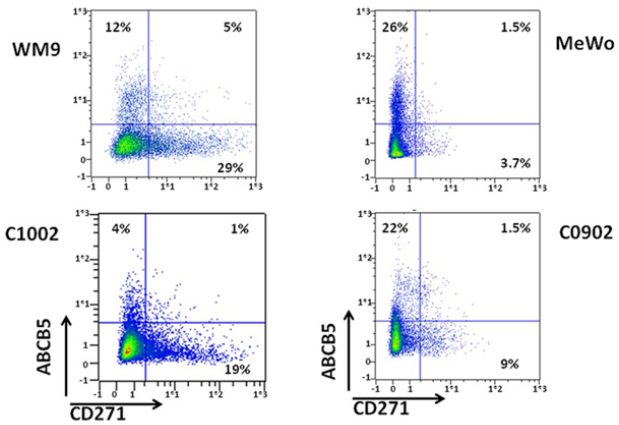
Shipping & Handling

Shipping Condition:	Dry Ice
Storage Condition:	Store Anti-ABCB5 at -20° C prior to opening. Aliquot antibody and freeze at -20° C or below for extended storage. Avoid cycles of freezing and thawing. Centrifuge product if not completely clear after standing at room temperature. This product is stable for several weeks at 4° C as an undiluted liquid. Dilute only prior to immediate use.
Expiration:	Expiration date is one (1) year from date of receipt.

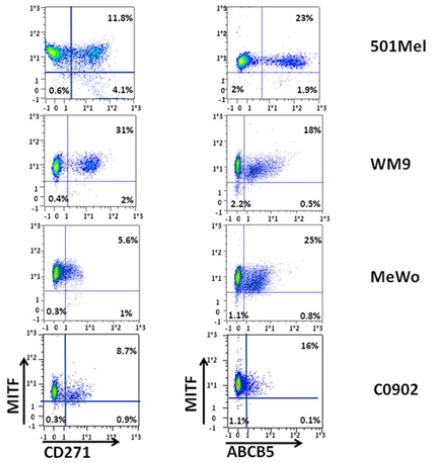
Images



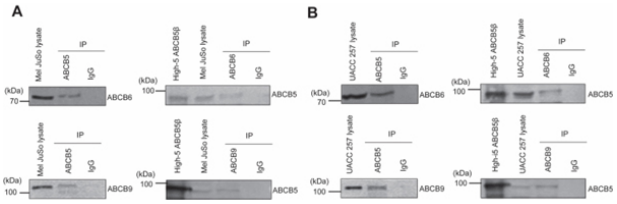
Western Blot
(C) Immunoblot showing that protein expression of ABCB5 is decreased in WM3734 cells transduced with TNC shRNA (sh_TNC_A and sh_TNC_C) when compared to non-targeting shRNA (sh_Cont). β-actin indicates equal loading. Fig 5. PMID: 20729912



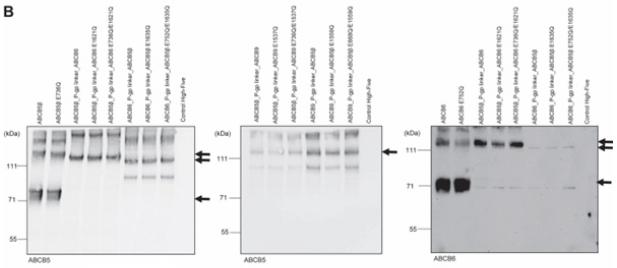
Flow Cytometry
Analysis of ABCB5 and CD271 populations in melanomas. WM9, MeWo, C1002 and C0902 were labeled for CD271 and ABCB5. Cells were analyzed by flow cytometry. CD271 intensity was plotted on abscissa and ABCB5 on ordinate. Fig 1. PMID: 25105565



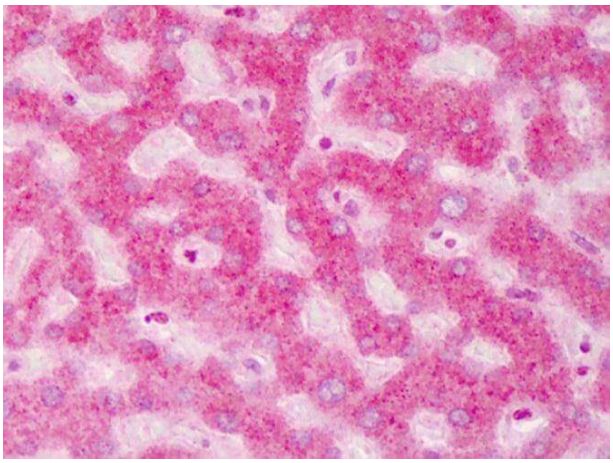
Flow Cytometry
Analysis of MITF expression in CD271 and ABCB5 populations.
WM9, 501-Mel, MeWo and C0902 were labeled for CD271 and MITF (left panel) or ABCB5 and MITF (right panel). CD271 or ABCB5 intensity was plotted on abscissa, and MITF expression on ordinate. Fig 2. PMID: 25105565



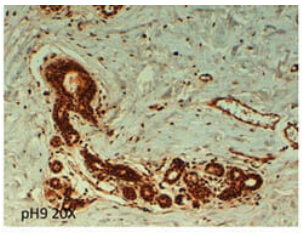
Immunoprecipitation
Coimmunoprecipitation of ABCB5β-ABCB6 and ABCB5β-ABCB9 demonstrates the presence of these heterodimers in Mel JuSo and UACC-257 cells. The Mel JuSo (A) or UACC-257 (B) proteins were immunoprecipitated (IP) with either an anti-ABCB5, anti-ABCB6, or anti-ABCB9 antibody. The precipitated proteins were revealed by Western blotting after SDS-PAGE using the corresponding antibody indicated on the right side of each blot. Fifteen micrograms of total proteins from the starting cell lysate were loaded in the first lane, while the total IP eluate was loaded on the gel. ABCB5β was expressed in High-Five insect cells, and total membrane proteins were prepared and loaded on the gel (High5 ABCB5β) as a complementary molecular weight marker when using anti-ABCB5. An isotype control was performed to determine the specificity of the signal obtained in Western blot. Fig 3. PMID: 38145744

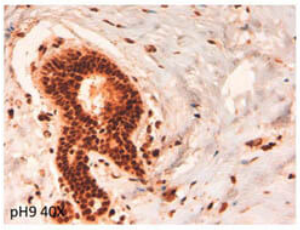


Western Blot
Expression of ABCB5β, ABCB6, ABCB5β_P-gp linker_ABCB6, ABCB6_P-gp linker_ABCB5β, ABCB9, ABCB5β_P-gp linker_ABCB9, and ABCB9_P-gp linker_ABCB5β in High-Five insect cells. Total membranes vesicles prepared from High-Five cells infected with baculovirus containing either ABCB5β, ABCB6, ABCB9, ABCB5β_P-gp linker_ABCB6, ABCB6_P-gp linker_ABCB5β, ABCB5β_P-gp linker_ABCB9, ABCB9_P-gp linker_ABCB5β and their corresponding EQ mutants were subjected to SDS-PAGE, followed by Western blotting with anti-ABCB5 and anti-ABCB6 antibodies (B). Bands of interest are highlighted by black arrows. Fig 5. PMID: 38145744

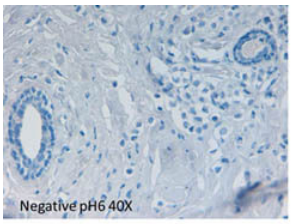


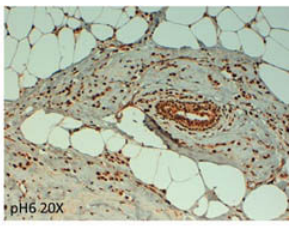
Immunohistochemistry
Immunohistochemistry of ABCB5 antibody. Tissue: human liver. Fixation: formalin fixed paraffin embedded. Antigen retrieval: user optimized. Primary antibody: ABCB5 at 1:200. Secondary antibody: Peroxidase goat anti-rabbit at 1:10,000 for 45 min at RT. Localization: Moderate to strong cytoplasmic and membranous staining was observed in hepatocytes. Occasional nuclear staining was observed in hepatocytes and sinusoidal cells. Staining: antibody as precipitated red signal with a hematoxylin purple nuclear counterstain.

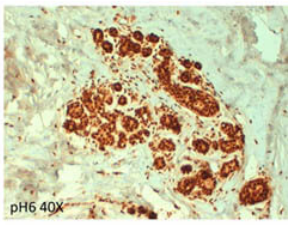




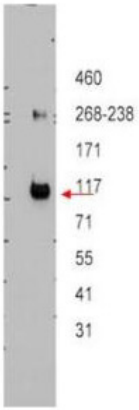
Immunohistochemistry
Immunohistochemistry of Rabbit anti ABCB5 antibody. Tissue: human breast carcinoma at pH9. (20X, 40X) Fixation: formalin fixed paraffin embedded. Primary antibody: ABCB5 antibody at 10 µg/mL for 1 h at RT. Secondary antibody: Peroxidase rabbit secondary antibody at 1:10,000 for 45 min at RT. Localization: ABCB5 is cytoplasmic. Staining: ABCB5 as precipitated brown signal.







Immunohistochemistry
Immunohistochemistry of Rabbit anti ABCB5 antibody. Tissue: human breast carcinoma at pH6. (left, neg control) (right, 20X, 40X). Fixation: formalin fixed paraffin embedded. Primary antibody: ABCB5 antibody at 10 µg/mL for 1 h at RT. Secondary antibody: Peroxidase rabbit secondary antibody at 1:10,000 for 45 min at RT. Localization: ABCB5 is cytoplasmic. Staining: ABCB5 as precipitated brown signal with hematoxylin purple nuclear counterstain.



Western Blot
Western blot using Rockland's affinity purified anti-ABCB5 antibody shows detection of ABCB5 beta in ~12.5 ug of transfected-Hi5 whole cell lysate. No reaction was seen when antibody was pre-incubated with the immunizing peptide (data not shown). A 3-8% Tris-acetate gel was used for separation. The arrowhead corresponds to 117 kDa ABCB5. The membrane was probed with the primary antibody at a 1:10,000 dilution in 5% milk in TBST at 4° C, overnight. Personal Communication, JP Gillet, CCR-NCI, Bethesda, MD.

References

- Gerard L et al. Identification of two novel heterodimeric ABC transporters in melanoma: ABCB5β/B6 and ABCB5β/B9. *J Biol Chem.* (2023)
- Cheli, Y et al. CD271 is an imperfect marker for melanoma initiating cells. *Oncotarget* (2014)
- Fukunaga-Kalabis M et al. Tenascin-C promotes melanoma progression by maintaining the ABCB5-positive side population. *Oncogene.* (2010)

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